

## CRYPTOSPORIDIUM INFECTION IN HUMAN DIARRHOEA PATIENTS IN WEST AZERBAIJAN, IRAN

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### ABSTRACT

The stool samples from 363 diarrhoeic and non-diarrhoeic children and adults from several locations in two towns of west Azerbaijan area of Iran examined for *Cryptosporidium* infection by modified Ziehl-Neelsen technique revealed the presence of infection in 7.66% of diarrhoeic patients and in none of the non-diarrhoeic individuals. The percentage of detection was higher among females (11.21%) than males (4.96%). 10.12 percent of children below five years of age and 4.65% of those between five and 15 years of age were positive, whereas only 2.12% of the adults were positive. The study has brought to light the existence of human *Cryptosporidium* infection in Iran perhaps for the first time.

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### INTRODUCTION

*Cryptosporidium*, a small coccidial parasite, though recognised 80 years ago, has remained until recently nothing more than a biomedical curiosity.<sup>1-3</sup> This protozoan enteropathogen is now regarded to be one of the most common causes of gastroenteritis and diarrhoea in man (especially children) and several animal species.<sup>4,5</sup> The first human cases of *Cryptosporidium* causing acute enterocolitis were reported in 1976.<sup>4,7</sup> Since then it has been found as the most common significant cause of diarrhoeal illness in various studies among human populations in different parts of the world.<sup>8-10</sup> Subsequent reports have shown that *Cryptosporidium* can also produce a short-term diarrhoeal illness in immunocompetent persons and severe diarrhoea in immunocompromised patients, especially those with the acquired immune deficiency syndrome (AIDS).<sup>11-13</sup>

Though diarrhoea and enteric diseases are not uncommon in Iran, there seems to be no report on the incidence of *Cryptosporidium* infection in human patients of this country. The present study was conducted

in two towns of west Azerbaijan area of Iran to determine the possible incidence of *Cryptosporidium* in diarrhoeic patients and healthy individuals.

### MATERIALS AND METHODS

The study was conducted in Urmia and Naghadeh towns of west Azerbaijan area in north west Iran from 23rd October 1988 to 22nd June 1989, and from 6th September 1989 to 4th February 1990, respectively. The towns of Urmia and Naghadeh are situated at a distance of about 950 kms from the capital Tehran and about 100 kms from each other. A total of 363 human samples (248 from diarrhoeic patients and the rest from nondiarrhoeic individuals) were collected, one from each individual, and examined within two hours of collection without any storage or significant transportation. The consistency and physical characteristics of each sample were recorded along with the health status and other specifications of the individual from whom the sample was collected. The samples were examined after processing and staining by modified

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**Table 1. Details of stool samples collected from diarrhoeic and non-diarrhoeic individuals from various locations of West Azarbaijan area of Iran**

| Locations<br>Urmia                      | Number of samples collected |    |                 |    |                  |    |                |   |                 |    |                  |    | Total of<br>diarrhoeic and<br>non-diarrhoeic |
|---|-----------------------------|----|-----------------|----|------------------|----|----------------|---|-----------------|----|------------------|----|--|
|   | DIARRHOEIC                  |    |                 |    |                  |    | NON-DIARRHOEIC |   |                 |    |                  |    |  |
|   | Group I<br>* +<br>M F       |    | Group II<br>M F |    | Group III<br>M F |    | Group I<br>M F |   | Group II<br>M F |    | Group III<br>M F |    |  |
| Shahid Gholipuri<br>Pediatrics Hospital | 31                          | 19 | -               | -  | -                | -  | -              | - | -               | -  | -                | -  | 50   |
| Taleghani Hospital                      | -                           | -  | -               | -  | 4                | 3  | -              | - | -               | -  | -                | -  | 7  |
| Urmia pathobiology<br>Laboratory        | -                           | -  | 3               | 2  | 5                | 10 | -              | - | -               | -  | -                | -  | 20   |
| Urmia Hygiene Center                    | -                           | -  | 2               | 2  | 6                | 3  | -              | - | -               | -  | -                | -  | 13   |
| College of Veterinary<br>Medicine       | -                           | -  | -               | -  | -                | -  | -              | - | -               | 50 | -                | -  | 50   |
| Total Urmia                             | 31                          | 19 | 5               | 4  | 15               | 16 | -              | - | -               | -  | 50               | -  | 140  |
| Naghadeh<br>Imam Hospital               | 6                           | 2  | 12              | 14 | 8                | 8  | -              | - | -               | -  | 15               | 16 | 81   |
| Naghadeh emergency<br>center            | 34                          | 20 | 4               | 4  | -                | -  | 4              | 5 | -               | -  | 13               | 6  | 90   |
| Private pediatrics<br>office            | 26                          | 20 | -               | -  | -                | -  | 3              | 3 | -               | -  | -                | -  | 52   |
| Total Naghadeh                          | 66                          | 42 | 16              | 18 | 8                | 8  | 7              | 8 | -               | -  | 28               | 22 | 223  |
| total Urmia                             | 67                          | 61 | 21              | 22 | 23               | 24 | 7              | 8 | -               | -  | 78               | 22 | 363  |

\* Male

Note : No sample could be collected from non-diarrhoeic individuals of Group II

+ Female

Ziehl-Neelsen technique.<sup>14</sup> The samples collected were grouped in three categories in accordance with the age of individuals from whom they were collected as follows:

Group-I: children less than 5 years of age

Group-II: children between 5 and 15 years of age

Group-III: adults (above 18 years of age).

The details of the collection of samples are presented in Table I.

### RESULTS

None of the 115 samples collected from the nondiarrhoeic individuals (including male and female adults and children) revealed the presence of *Cryptosporidium* oocysts. Thus no *Cryptosporidium* infection was discovered in non-diarrhoeic humans either in Urmia or in Naghadeh. However, of the 248 samples collected from diarrhoeic individuals of the study, 19 (7.66%) were positive for *Cryptosporidium* (Table II.). Of the 19 positive cases, seven were males and 12 females, sixteen were from group-I, two from group-II and one from group-III. Thus seven out of 141 diarrhoeic males (4.96%) and 12 out of 107 diarrhoeic females (11.2%) were positive. Sixteen out of the 158 children of group-I (10.12%) and two out of 43 children of group-I and group-II (4.65%) were positive. Taking all the

children of group-I and group-II together, 18 out of 201 (8.95%) were positive, whereas only one out of 47 diarrhoeic adults of group-III (2.12%) was positive.

A comparison of the results of Urmia and Naghadeh shows that the latter town had a higher overall percentage of positive cases (8.23% against 6.67% in Urmia). The positive cases of group-I were approximately the same proportion in both towns (10% and 10.18%), respectively. However in Naghadeh two out of 34 children of group-II (5.89%) were also positive, whereas none of the children of this group tested in Urmia was positive. In Naghadeh again, 3.23% diarrhoeic females were positive whereas in Urmia the corresponding percentage was 7.69.

### DISCUSSION

The present study revealed the hitherto unreported existence of *Cryptosporidium* infection among humans with diarrhoea in West Azerbaijan area of Iran. It is likely that the disease may also be prevalent in the remaining areas of Iran. The fact that none of the non-diarrhoeic individuals tested in same areas where disease was recognised in diarrhoeic individuals excreted oocysts, shows the rarity or possible absence of subclinical or non-clinical forms of the disease in these areas. A 3% asymptomatic infection rate in rural

Table II. Details of samples positive for Cryptosporidiosis in Urmia and Naghadeh towns of West Azerbaijan area of Iran

| Locations<br>Urmia                | Number of Samples |                     |                   |                     |                   |                     | Number<br>Total | Number<br>Positive |
|-----------------------------------|-------------------|---------------------|-------------------|---------------------|-------------------|---------------------|-----------------|--------------------|
|                                   | Group I           |                     | Group II          |                     | Group III         |                     |                 |                    |
|                                   | Samples<br>tested | samples<br>positive | samples<br>tested | samples<br>positive | samples<br>tested | samples<br>positive |                 |                    |
| Male                              | 31                | 3                   | 5                 | 0                   | 15                | 0                   | 51              | 3                  |
| Female                            | 19                | 2                   | 4                 | 0                   | 16                | 1                   | 39              | 3                  |
| Total                             | 50                | 5                   | 9                 | 0                   | 31                | 1                   | 90              | 6                  |
| Naghadeh                          |                   |                     |                   |                     |                   |                     |                 |                    |
| Male                              | 66                | 3                   | 16                | 1                   | 8                 | 0                   | 90              | 4                  |
| Female                            | 42                | 8                   | 18                | 1                   | 8                 | 0                   | 68              | 9                  |
| Total                             | 108               | 11                  | 34                | 2                   | 16                | 0                   | 158             | 13                 |
| Total of<br>Urmia and<br>Naghadeh | 158               | 16                  | 43                | 2                   | 47                | 1                   | 248             | 19                 |

populations has been reported.<sup>15</sup>

The overall percentage of diarrhoeic individuals positive for *Cryptosporidium* infection in this study (7.66%) was much lower than the figure of 21% reported by Kwage and coworkers<sup>16</sup> from Nigeria and higher than the corresponding figures of 1.4%, 1.2%, 4.1% and 5.6% reported from UK, Canada, Australia and India, respectively.<sup>17-20</sup> Epidemiological studies have indicated a prevalence of 1-5% amongst those with diarrhoea in developed countries.<sup>21</sup>

The results of this study have revealed that the prevalence of *Cryptosporidium* in diarrhoeic individuals was highest amongst children below the age of five years (10.12%), followed by children between five to 15 years of age (4.65%), and least amongst the adults (2.12%). The results of parasitological surveys also showed that children usually have a higher prevalence than adults.<sup>19,22,23</sup> Amongst the children below five years with gastroenteritis, Robinson, et al<sup>24</sup> and Shahid, et al<sup>25</sup> have reported a prevalence of 6.1% in Sudan and Bangladesh, respectively, while Hojlung, et al<sup>26</sup> have reported a prevalence of 7.9% in the same age group from Liberia. Tzipori and coworkers<sup>19</sup> have reported that in Australia the prevalence of cryptosporidiosis among children with gastroenteritis was higher (4.8%) than that in adults (1.6%). In a waterborne outbreak of cryptosporidiosis in UK, 17 out of the 27 confirmed cases were children aged four months to eight years.<sup>27</sup> In Switzerland, 5.5% of children with diarrhoea identified by a laboratory based survey were found to be positive for oocysts of *Cryptosporidium* spp.<sup>28</sup> In the same country, 4.6% of children with diarrhoea who attended a hospital from June to September, 1988 were positive for *Cryptosporidium*.<sup>29</sup> However, contrary to these reports, results from Finland and Nigeria have indicated that adults were more frequently infected than children.<sup>15,16</sup> The reason for lower incidence amongst children in Finland is obscure

but may be due to breast feeding, which during the past few years has been intensively encouraged in that country.<sup>30</sup> The authors of the study from Nigeria have reported that the higher prevalence among adults may, in part, be due to few cases investigated and lower number of children included in their study.<sup>16</sup>

Another feature of our results was the detection of a higher percentage of *Cryptosporidium* amongst females (11.21%) as compared to that in males (4.96%). Seventeen of the 27 (62.9%) confirmed cases of cryptosporidiosis observed by Smith and coworkers<sup>27</sup> in a waterborne outbreak of this disease in Ayrshire U.K. were females. The authors have stated that the reason for this higher proportion of females than males (1.7:1) were "unknown" and have also cited a similar higher attack rate in females observed earlier by Hayes and colleagues.<sup>31</sup>

Interest in *Cryptosporidium* spp. by the veterinary medical profession has considerably increased for the past two decades after the first publication of a case in a calf by Panciera and coworkers in 1971.<sup>32</sup> Cryptosporidiosis has been considered as an "emerging zoonosis"<sup>33</sup> and numerous publications reporting the infection of various species of animals and birds with *Cryptosporidium* spp. and a possible link between these and human infections are now available.<sup>5,9,28</sup> More studies are however needed on the epidemiology of *Cryptosporidium* infection in man and animals with particular reference to interspecies infectivity. Some of these investigations by the present authors are now underway in Iran and the results will be reported in due time.

At the time of writing we are not aware of any published evidence of prevalence of *Cryptosporidium* infection in human diarrhoea patients in Iran. This may therefore be the first report showing the existence of this parasite in the West Azerbaijan area of this country.

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